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Copper dosing to biological rapid sand filters increases nitrifier activity and abundance

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Introduction

Drinking water quality can deteriorate when nitrification is incomplete during treatment with biological rapid sand filters. Poor nitrification performance can be caused by limited availability of nutrients, required for the nitrifying microorganisms. The micronutrient copper for example is specifically required for the enzyme ammonia monooxygenase, mediating ammonia oxidation in ammonia oxidizing bacteria (Sayavedra-Soto and Arp, 2011).

Here, we hypothesized that deficiency of copper could hamper nitrification in biological filters treating groundwater, and that limitations could be removed by addition of copper. We furthermore assumed that a possible increase in nitrification activity would be associated with a change in nitrifier abundance. To test the above, we studied the effect of copper dosing on nitrification activity in a full-scale biological rapid sand filter treating groundwater, and investigated whether the dosing would affect the abundance of nitrifying microorganisms. Obtained information can be indicative of the possibility of engineering microbial communities in biofilters through addition of specific nutrients.

Material and Methods

Copper at up to 5 $\mu\text{g Cu L}^{-1}$ was dosed for 117 days employing an electrolysis system (patent pending) to a filter at a Danish water treatment plant with a long history of poor nitrification performance. To closely follow the effect of copper dosing on nitrification, influent and effluent ammonium concentrations were monitored with 30 minute frequency using an ammonium auto analyser (Hach Lange, AMTAXTM sc). (Wagner et al., 2016)

Filter media core samples (0-40 cm filter depth) were collected one day before dosing started, and 22 and 116 days with copper dosing. Core samples were subdivided into 10 cm long sections and DNA was extracted (MP FastDNATM SPIN Kit for soil) from approx. 0.5 g (drained wet weight) of each subsample. Content of total Eubacteria and ammonia oxidizing bacteria (AOB) was quantified through qPCR analysis, targeting the specific regions on the 16S rRNA gene. Primers used were 1055f and 1392r for total Eubacteria, and CTO 189fA/B/C and RT1r for AOB (*beta-Proteobacteria*). Gene copy numbers from qPCR analysis were converted to cell copy numbers, assuming one 16S rRNA gene copy per cell.

Results and Conclusions

Copper dosing to the full-scale filter stimulated ammonium removal within one day, and decreased effluent concentrations from approx. 0.18 to < 0.02 mg $\text{NH}_4\text{-N L}^{-1}$ within 20 days (Fig. 1 A). Water sampling over depth of the filter revealed that ammonium oxidation activity moved upwards in the filter with dosing onset. Thereby, stratified removal was created, and

the ammonium removal rates of upper layers of the filter increased significantly. E.g., within 57 days of dosing, the ammonium removal rate of the top 10 cm of the filter increased almost 14-fold. A control filter, without copper dosing but equal ammonium loading, showed no change in ammonium removal performance. (Wagner et al., 2016)

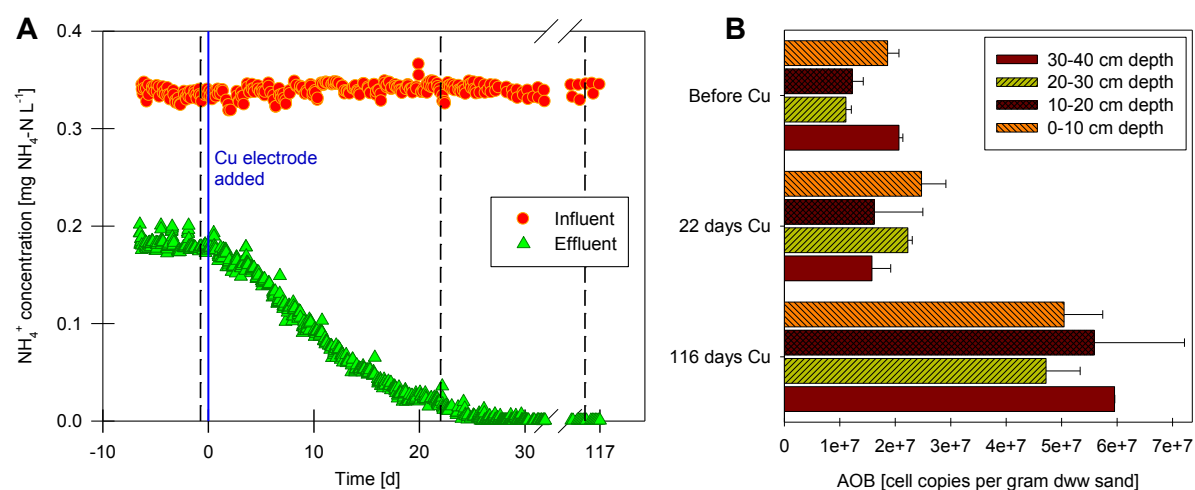


Figure 1 - (A) Influent and effluent ammonium concentrations of the full-scale filter. Copper dosing was started on day 0. The dashed lines denote sampling of filter material one day before, and 22 and 116 days with copper dosing. Fig. 1 A is adapted from Wagner et al. (2016). (B) AOB gene copy numbers per gram drained wet weight [dww] of sand, from 0-40 cm depth, for the three filter media sampling days. Error bars give the standard deviation from results of triplicate qPCR analysis.

Abundance of AOB in the top 40 cm of the filter was only marginally increased after 22 days of copper dosing, but tripled within 116 days (Fig. 1 B). The number of total Eubacteria did not change over time with dosing (data not shown); hence, the relative abundance of AOB increased. Before dosing, the fraction of AOB to Eubacteria in the top 40 cm of the filter was 0.8 %. This ratio is comparable to previous findings from other biological sand filters treating groundwater (Lee et al., 2014; with same qPCR primers used). Copper dosing increased the fraction of AOB to Eubacteria to 1.0 % after 22 days, and further to 3.0 % after 116 days of dosing. Our results show that the copper induced stimulation of ammonium removal is not only caused by a higher (enzymatic) activity of an existing population of AOB, but that with time, also their abundance is positively affected.

Overall, the study shows that copper dosing at low concentrations can be a powerful tool for improving nitrification performance of biological filters. Furthermore, it demonstrates that (selective) nutrient dosing can be used to engineer microbial communities to improve biological drinking water treatment. The research can therefore have a great impact on the water treatment industry.

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